

Amendment to the Claims:

Claims 1-50 (Canceled)

51. (Currently amended) A method of producing a targeting construct, the method comprising:

- (a) amplifying a polynucleotide sequence comprising a first ~~region sequence~~ homologous to a first region of a target gene or sequence and a second ~~region sequence~~ homologous to a second region of said target gene or sequence;
 - (b) providing a gene encoding a positive selection marker; and
 - (c) annealing the gene encoding the positive selection marker with the polynucleotide sequence to form the construct using ligation independent cloning, such that the gene encoding the positive selection marker is positioned between the first ~~region~~ homologous sequence and the second ~~homologous sequence~~ region of the target gene or sequence;
- wherein the first ~~region sequence~~ and second ~~sequence~~ region are capable of homologously ~~recombine~~ recombining with the target gene or sequence.

52. (Previously presented) The method of claim 51, wherein the polynucleotide sequence is amplified directly from a plasmid library.

53. (Currently amended) The method of claim 51, wherein the gene encoding the positive selection marker is a neomycin resistance gene.

54. (Currently amended) The method of claim 51, wherein the polynucleotide sequence is amplified with oligonucleotide primers each comprising a 5' terminal sequence consisting of that are at least 12-5 nucleotides selected from no more than three of the following types of nucleotides: A, T, C and G in length and have a 5' sequence lacking one type of base.

55. (Currently amended) The method of claim 51, wherein the construct further comprises a gene encoding a screening marker.

56. (Previously presented) The method of claim 55, wherein the screening marker is a fluorescent protein.

57. (Currently amended) A method of producing a targeting construct, the method comprising:

- (a) providing a circular plasmid library comprising nucleotide fragments corresponding to a target gene sequence;
- (b) amplifying a ~~first~~ polynucleotide sequence comprising a first sequence homologous to a first region of said target gene sequence and a second sequence homologous to a

~~second region of said target gene sequence~~~~corresponding to a target gene or sequence~~
~~from a circular plasmid library, wherein the polynucleotide sequence comprises first~~
~~and second regions which are homologous to the target gene or sequence; and~~
~~(b)(c) inserting a second polynucleotide sequence~~gene encoding a positive selection
marker between the first region sequence and the second region sequence to form the
construct using ligation independent cloning,
wherein the first region sequence and the second region are capable of homologously
~~recombine-recombining~~ with the target gene or sequence.

58. (Currently amended) The method of claim 57, wherein the gene encoding the positive
selection marker is a neomycin resistance gene.
59. (Currently amended) The method of claim 57, wherein the polynucleotide sequence is
amplified with oligonucleotide primers each comprising a 5' terminal sequence consisting of
~~that are at least 12-5 nucleotides in length selected from no more than three of the following~~
types of nucleotides: A, T, C and G and have a 5' sequence lacking one type of base.
60. (Currently amended) The method of claim 57, wherein the construct further comprises a gene
encoding a screening marker.
61. (Previously presented) The method of claim 60, wherein the screening marker is a
fluorescent protein.
62. (New) The method of claim 54, wherein the 5' terminal sequence consists of at least 12
nucleotides selected from no more than three of the following types of nucleotides: A, T, C
and G.
63. (New) The method of claim 62, wherein the 5' terminal sequence consists of at least 20
nucleotides selected from no more than three of the following types of nucleotides: A, T, C
and G.
64. (New) The method of claim 59, wherein the 5' terminal sequence consists of at least 12
nucleotides selected from no more than three of the following types of nucleotides: A, T, C
and G.
65. (New) The method of claim 64, wherein the 5' terminal sequence consists of at least 20
nucleotides selected from no more than three of the following types of nucleotides: A, T, C
and G.